

Robertson-Andersson, D. V., et al. (2011). Effects of wild and farm-grown macroalgae on the growth of juvenile South African abalone *Haliotis midae* Linnaeus. AFRICAN JOURNAL OF AQUATIC SCIENCE, 36(3):331-337



Effects of wild and farm-grown macroalgae on the growth of juvenile South African abalone *Haliotis midae* Linnaeus

D.V. Robertson-Andersson, G.W. Maneveldt* and K. Naidoo

Department of Biodiversity and Conservation Biology, University of the Western Cape, Private Bag X17, Bellville 7535, South Africa.

*Corresponding author: gmaneveldt@uwc.ac.za

Abstract

The effect of various macroalgal diets on the growth of grow-out (> 20 mm shell length) South African abalone *Haliotis midae* was investigated on a commercial abalone farm. The experiment consisted of four treatments: fresh kelp blades (*Ecklonia maxima* [Osbeck] Papenfuss) (~ 10 % protein); farmed, protein-enriched *Ulva lactuca* Linnaeus (~26 % protein) grown in aquaculture effluent; wild *U. lactuca* (~ 20 % protein); and a combination (mixed) diet of kelp blades + farmed *U. lactuca*. Abalone grew best on the combination diet ($0.423 \pm 0.02\%$ weight day⁻¹ SGR [specific growth rate]; $59.593 \pm 0.02 \mu\text{m day}^{-1}$ DISL [daily increment in shell length]; 1.093 final CF [condition factor]) followed by the kelp only diet ($0.367 \pm 0.02 \%$ weight day⁻¹ SGR; $53.148 \pm 0.02 \mu\text{m day}^{-1}$ DISL; 1.047 final CF), then the farmed, protein-enriched *U. lactuca* only diet ($0.290 \pm 0.02\%$ weight day⁻¹ SGR; $42.988 \pm 0.03 \mu\text{m day}^{-1}$ DISL; 1.013 final CF) that in turn outperformed the wild *U. lactuca* only diet ($-0.079 \pm 0.01 \%$ weight day⁻¹ SGR; $3.745 \pm 0.02 \mu\text{m day}^{-1}$ DISL; 0.812 final CF). The results suggest that protein alone could not have accounted for the differences produced by the varieties of *U. lactuca* and that the gross energy content is probably important.

Key words: abalone, *Ecklonia maxima*, farm-grown, growth, *Haliotis midae*, mixed diet, protein content, *Ulva lactuca*

Introduction

Kelp is the major macroalgal feed for farmed abalone in South Africa (Troell et al. 2006). With the expansion of the abalone industry in recent years, the demand for fresh kelp has steadily grown (Anderson et al. 2006, Rothman et al. 2006, Francis et al. 2008a). In 2003, more than 7000 tons of fresh kelp fronds were harvested (Robertson-Andersson et al. 2008). By 2009, however, this demand dropped to about 5500 tons (Marine and Coastal Management, South African Department of Agriculture, Forestry and Fisheries, unpublished data), this largely due to the

incorporation of formulated feeds into abalone diets over the past few years (P. Britz, Department of Ichthyology and Fisheries Science, Rhodes University, *pers. comm.*). Despite this reduction in kelp for feed, many kelp beds were still approaching limits of sustainable harvesting, particularly in kelp concession areas with high numbers of abalone farms (Anderson et al. 2003, 2006, Rothman et al. 2006, Troell et al. 2006).

There are other drawbacks to the use of kelp as a fresh feed. While kelp is cheaper than formulated feeds on a price per ton basis, its future availability as the primary abalone fresh feed is uncertain (Troell et al. 2006). Kelp harvesting is also dependent on sea conditions; this complicates farm management and increases the risk of such ventures (Dixon 1992, Britz 1995). The use of kelp was considered by some farmers to be costly and labour intensive when the expenses of harvesting, transport and storage were taken in account (W. Barnes, Abalone Farmers Association of South Africa [AFASA], *pers. comm.*). Other, less desirable properties of kelp include its low protein (5 - 15 %) and high water (68 - 83 %) content (Hahn 1989), and its unbalanced amino acid profile (Britz 1996a, Erasmus et al. 1997, Rosen et al. 2000, Troell et al. 2006). As an alternative to kelp, some South African farms already grow certain species of macroalgae for use as abalone feed in their aquaculture systems.

Farm-grown macroalgae have been shown to have comparatively higher protein contents than their wild counterparts (Neori and Shpigel 1999, Shpigel et al. 1999, Neori et al. 2003, 2004, Robertson-Andersson et al. 2008). This is because macroalgae remove dissolved nitrogen from aquaculture effluent resulting in a considerable increase in the alga's phosphorous and protein content (Robertson-Andersson et al. 2008). Of all macroalgae grown in aquaculture environments, *Ulva* spp. have been shown to be the best candidates for integrated aquaculture practices largely because they are fast-growing, have a high nitrogen concentration, are generally resistant to epiphytes, and have simple life cycles that are easily controlled (Shpigel et al. 1996, Neori and Shpigel 1999, Shpigel et al. 1999, Neori et al. 2003, 2004).

The cultivation of *Ulva* spp. on South African abalone farms is increasing, with two such farms depending almost entirely on farm-grown macroalgae to meet the bulk of their feed requirements (Bolton 2006, Troell et al. 2006, Robertson-Andersson et al. 2008). The latter farms culture *Ulva lactuca* Linnaeus alongside *Haliotis midae* in an integrated mariculture system where the algae serve not only as a biological filter, but also as feed for the abalone (Bolton 2006, Troell et al. 2006, Robertson-Andersson et al. 2008, Bolton et al. 2009). While data on *H. midae* feed fortification with wild macroalgae exist (e.g. Stepto and Cook 1993, Naidoo et al. 2006, Dlaza et al. 2008), information on how *H. midae* may benefit from farm-grown, protein-enriched macroalgae is lacking. The objective of this study was therefore to compare the growth of juvenile grow-out *H. midae* on both farm-grown and wild *U. lactuca*, and study the effects of wild macroalgal feed fortification with the protein-enriched, farm-grown *U. lactuca*.

Materials and methods

Experimental system

The research was conducted at the Jacobsbaai Sea Products (JSP - 17° 53' 12.5" E, 32° 58' 2.5" S, Western Cape, South Africa) commercial abalone farm. Moderately aerated seawater flowing at $700 \pm 100 \text{ Lh}^{-1}$ was supplied at $15.5 \pm 2.5 \text{ }^{\circ}\text{C}$ in a flow-through concrete holding tank (L x B x H; 5500 x 1300 x 550mm) with a total water volume of 4500 L. Table 1 details water quality variables measured. Abalone were grown in polyethylene culture baskets (L x B x H; 1037 x 517 x 540mm) subdivided with four vertically-orientated ABS (acrylonitrile butadiene styrene) plates (L x B x H; 760 x 4 x 310 mm) to increase the surface area to approximately 1.8 m² in each basket available for attachment. A horizontal polyethylene feeder plate (L x B x H; 600 x 4 x 380mm) was positioned centrally above the vertical plates. This design provided optimum access to feed with no visible feed wastage. Replicate ($N = 4$) baskets were arranged in a single holding tank according to a Latin square model (Zar 1984). In addition, the flow direction in the tank was alternated weekly to further compensate for end effects. End effects refer to the potential negative impacts (e.g. reduced water flow-rate, accumulation of faeces and silt, etc) that may arise from a unidirectional flow. Both the flow-through tank and culture baskets were cleaned every two weeks.

Experimental animals

Hatchery-reared grow-out (abalone with a shell length > 20mm) abalone (supplied by the JSP commercial abalone farm), approximately 22 months old from a single brood stock pool, were subdivided into four diet treatments of approximately 2.5 kg (approximately 250 individuals) abalone per basket per diet treatment totaling 4000 animals used for the study. The stocking density in the tank was approximately 10 kg m⁻³. Initial shell length and body weight were $36.04 \pm 0.16 \text{ mm}$ and $8.69 \pm 0.12 \text{ g}$ respectively. After four months, stocking density of each basket was reduced by random removal of animals to 150 individuals to reduce feed competition.

Treatments

Four dietary treatments were tested, each with four replicate baskets:

1. Fresh kelp (*Ecklonia maxima* [Osbeck] Papenfuss) blades. Kelp was chosen as a seaweed control because it is this seaweed that is most commonly used as fresh abalone feed in South Africa.
2. Wild *U. lactuca* collected from the adjacent bay.
3. Farmed *U. lactuca* obtained from the JSP farm from a cultured stock grown in both abalone and fish (turbot, *Scophthalmus maximus* Linnaeus) effluent.
4. A mixed diet of fresh kelp blades and farmed *U. lactuca* supplied in equal quantities.

All feeds were spread evenly among the vertically-orientated plates and supplied daily *ad libitum* according to standard farm feeding practices with the total weight of

macroalgae not exceeding 150 g per basket per day. At each feeding, any leftover macroalgae were removed and fresh macroalgae added. Table 2 details the approximate nutritional composition of the diets.

Sampling and data collection

Representative animals were randomly selected for measurement from each basket at monthly intervals during the eight-month experiment ($n = 30$ at 0-2 months, $n = 40$ at 3-7 months and $n = 50$ at 8 months to compensate for increasing differential growth). Before measuring, abalone were blotted dry to remove excess water. Body weight was recorded to the nearest 0.01 g using an electronic balance, and shell length along the longest axis of the shell was measured to the nearest 0.01 mm with electronic Vernier calipers. Daily increment in shell length (DISL, $\mu\text{m day}^{-1}$) was calculated using the formula of Zhu et al. (2002): $\text{DISL} = [(\text{SL}_t - \text{SL}_i)/t] \times 1000$ where SL_t = final mean shell length (mm), SL_i = initial mean shell length (mm), and t = the feeding period in days.

Specific growth rate (SGR in % body weight day^{-1}) was calculated using the formula of Britz (1996b): $\text{SGR} = ([\ln(W_f) - \ln(W_i)]/t) \times 100$ where $\ln(W_f)$ = the natural log of the final mean weight, $\ln(W_i)$ = the natural log of the initial mean weight, and t = the feeding period in days. The condition factor (CF), which is an index developed to account for the relationship between the weight of the abalone per unit shell length, was calculated using the formula of Britz (1996b): $\text{CF} = [\text{BW} / \text{SL}^{2.99}] \times 5575$ where BW = the body weight (g) and SL = the shell length (mm).

Feed conversion ratio (FCR) was calculated using the formula of Britz (1996b): $\text{FCR} = \text{R}/\text{G}$ where R = ration, defined as the blotted wet feed intake (g), and G = growth, defined as the blotted wet weight (g) gained. To calculate FCR, the weight of all algae placed into the baskets was recorded and any uneaten algae that was removed from the baskets was recorded. Kelp was allowed to drip-dry in empty culture baskets for no more than 5 min, while all *U. lactuca* samples were simply blotted dry.

Statistical analysis

Treatment average values for each of the four replicate baskets were analysed by means of a one-way analysis of variance (ANOVA), which assumes equal variances and normality of residuals. To test for this, a Levene's test for homogeneity of variances and the Shapiro-Wilk test for normality were performed for data from each month. All underlying assumptions were met. Pair-wise comparisons between treatments were analysed using Tukey's test for multiple comparison of means. Differences amongst treatments were considered statistically significant at $p < 0.05$. Unless otherwise stated, data are expressed as means \pm SE.

Results

Abalone grew fastest ($\text{SGR} = 0.423 \pm 0.02 \text{ \% weight day}^{-1}$; $\text{DISL} = 59.593 \pm 0.02 \text{ }\mu\text{m day}^{-1}$) on a combination diet of kelp and farm-grown, protein-enriched *U. lactuca* (weight ANOVA: $F_{(3,12)} = 387.29$; $p < 0.001$) (length ANOVA: $F_{(3,12)} = 542.23$; $p < 0.001$) (Figures 1 and 2, Table 3). Abalone fed farm-grown *Ulva lactuca* produced significantly better growth ($\text{SGR} = 0.290 \pm 0.02 \text{ \% weight day}^{-1}$; $\text{DISL} = 42.988 \pm 0.03 \text{ }\mu\text{m day}^{-1}$) than those fed wild *U. lactuca* ($\text{SGR} = -0.079 \pm 0.01 \text{ \% weight day}^{-1}$; $\text{DISL} = 3.745 \pm 0.02 \text{ }\mu\text{m day}^{-1}$) (weight T-test: $t = 66.32$; $p < 0.001$) (length T-test: $t = 205.61$; $p < 0.001$). Despite the improved protein content, farm-grown *U. lactuca* did not result in improved abalone growth compared with kelp as a single feed diet ($\text{SGR} = 0.367 \pm 0.02 \text{ \% weight day}^{-1}$; $\text{DISL} = 53.148 \pm 0.02 \text{ }\mu\text{m day}^{-1}$, $p < 0.001$) (weight T-test: $t = 22.43$; $p < 0.001$) (length T-test: $t = 10.63$; $p = 0.002$), which resulted in superior growth.

While all animals had CF values >1 at the start of the experiment, only abalone cultured on a mixed diet of kelp and farm-grown *U. lactuca* improved their CF values, producing comparatively ‘fatter’ individuals (Table 2). The remaining diets resulted in a reduction of CF values (Table 3) suggesting that abalone grown on these feeds gained more length relative to weight. This trend was particularly strong in the wild *U. lactuca* that showed a negative specific growth rate. CF ranking for all diets followed those of the SGR and DISL. Similarly as before, although farm-grown *U. lactuca* had higher protein contents than kelp, FCR for this alga was higher and thus feed conversion efficiency (FCE) lower, i.e. substantially more protein-enriched *U. lactuca* was required to produce comparable growth.

Discussion

While many studies (Owen et al. 1984, Day and Fleming 1992, Fleming 1995a, Simpson and Cook 1998, Naidoo et al. 2006, Dlaza et al. 2008) have shown the benefits of variability in the diets (e.g. mixed and rotational diets) of abalone, no studies exist to show the benefit of feeding farm-grown macroalgae over that of their wild counterparts. This study is the first to show that farm-grown *U. lactuca*, whether fed as a single diet or as part of a mixed diet, produced comparatively good growth in farmed *H. midae*. This was likely due to the increased protein content of the macroalga as they were farmed under aquaculture conditions.

Farm-grown, protein-enriched *U. lactuca* had produced good growth in abalone species such as *H. tuberculata* Linnaeus (Neori et al. 1998, Shpigel et al. 1999) and *H. discus hannai* Ino (Shpigel and Neori 1996, Neori and Shpigel 1999, Shpigel et al. 1999). Wild macroalgae on the other hand, are known to have a comparatively low protein content ranging from 4 - 20 % (Mercer et al. 1993, Fleming et al. 1996, Fleurence et al. 1999, Neori and Shpigel 1999, Shpigel et al. 1999, Tahlil and Juinio-Menez 1999, Rosen et al. 2000, Bautista-Teruel et al. 2002, Demetropoulos and Langdon 2004a, 2004b, Dlaza et al. 2008, Robertson-Andersson et al. 2008) that result in relatively slow growth in abalone (Fleming 1995b, Fleming et al. 1996, Neori and Shpigel 1999, Shpigel et al. 1999, Rosen et al. 2000, Kruatrachue et al. 2004, Lee

2004, Neori et al. 2004). Previous growth studies on *H. midae* (Cook and Claydon 1991, Owen et al. 1984, Stepto and Cook 1993, Simpson and Cook 1998) that used macroalgae as feed, have used only wild, relatively low-protein macroalgae that whether fed as single-species, or in combination as mixed diets, have never been able to produce growth comparable to that obtained with feeds relatively high in crude protein (Viana et al. 1993, Fleming 1995a,b, Fleming et al. 1996, Bautista-Teruel and Millamena 1999, Bautista-Teruel et al. 2002, Kruatachue et al. 2004, Lee 2004). To overcome this deficiency, Shpigel et al. (1999) suggested that it would be necessary to feed protein-enriched *U. lactuca* together with wild macroalgae as part of a mixed diet in order to achieve commercially acceptable growth rates. This was suggested because mixed diets essentially compensate for nutrients and attractants that are generally lacking in single-species diets.

Research into abalone protein requirements gave optimal protein levels of 36 % for *H. midae* (Sales et al. 2003a, 2003b). Although studying larger abalone of an initial size of around 50 mm shell length, Green et al. (2011) showed that the protein content of formulated feed for *H. midae* can be dropped as low as 18 % given that the digestible energy of the feed is not lower than 13.5 MJ kg⁻¹. The digestible energy content of food is that portion of the energy that is available to the animal after taking into account the energy costs associated with excretion i.e. that portion of the feed gross energy available to the body after the energy costs from excretion have been removed (Fleming 1995b, Britz and Hecht 1997). In *H. midae*, energy loss in the form of excretion has been recorded to range from being negligible (accounting for < 1 % of consumption; Barkai and Griffiths 1988), to a maximum of 15 % (Green et al. 2011; determined from Table 1). Thus, the digestible energy from feed available to *H. midae* can be assumed to be approximately equal to, or very close to the gross energy content. Bearing this variability in mind, and since Green et al. (2011) did not test diets with energy levels between 13.5 and 11.6 MJ kg⁻¹, we would argue that the farmed *U. lactuca* probably met the minimum energy requirements for optimal *H. midae* growth.

The protein contents of wild and farmed *U. lactuca* used in this study both meet the minimum percentage requirement proposed by Green et al. (2011), suggesting that protein alone could not have accounted for the differences observed in the growth rates experienced from these diets. To account for these very different growth rates, we propose two, but not necessarily mutually exclusive, scenarios. Firstly, both the protein and energy contents of the farmed *U. lactuca* were within the suggested “optimal range” and we suggest that the increased energy content of the farmed *U. lactuca* probably accounted for the marked differences observed between the wild and farmed varieties of the alga. Secondly, the fact that the abalone fed wild *U. lactuca* lost weight over the experimental period suggests that wild *U. lactuca* were unsuitable as a feed source. This being the case, any number of factors, e.g., amino acid profile, presence/absence of specific bacterial coatings, lack of attractants, toughness of the thallus, etc., may have contributed to the poor growth rates of abalone fed the wild *U. lactuca*.

While the results of this study are consistent with previous studies, there is one exception. Of the single-species diets tested, fresh kelp out-performed even the protein-enriched *U. lactuca*. This was surprising as both the protein and gross energy contents for kelp were significantly less than that recorded for the farm-grown *U. lactuca*. Furthermore, the kelp-only feed produced a FCR considerably lower (and thus higher feed conversion efficiency) than the farm-grown *U. lactuca* i.e. substantially more farm-grown *U. lactuca* was required to produce comparable growth. Despite this better performance, kelp in this study performed considerably poorer than that achieved in the South African abalone industry (Hattingh 2006) where FCR values ranging from 9.20 - 15.40 have been reported. Equally surprising is that the wild *U. lactuca* and kelp performed so differently, particularly as energy contents were very similar and protein content was higher in the wild *U. lactuca*. Theoretically, both wild and farmed *U. lactuca* should have performed better than kelp. The better performance of fresh kelp as a single feed diet might be explained by one of three possible reasons, or a combination thereof.

1. The diet prior to the start of the experiment was fresh kelp alone. This may explain the comparatively poor performance of the farmed *U. lactuca* as abalone in this size range often become habituated to kelp and are slow to change (P. Britz, *pers. comm.*).
2. The contribution of bacteria associated with fresh kelp may have added to the digestibility and assimilation of nutrients in the kelp (P. Britz in Troell et al. 2006: 274).
3. For some time, only protein contents and amino acid profiles of feeds were considered limiting factors (Britz 1996a, b, Fleming et al. 1996b, Britz and Hecht 1997, Troell et al. 2006). Research by Green et al. (2011) showed that digestible energy is also important. It is thus possible that some other, as yet unidentified variable or combination of variables, may also account for such differences.

Ulva lactuca has long been known to be a good candidate for aquaculture. This species has a high nutrient uptake capacity (Neori et al. 1998, 2004, Neori and Shpigel 1999, Shpigel et al. 1999), a rapid growth rate (Neori et al. 1996, Shpigel et al. 1999, Robertson-Andersson et al. 2006), and a relatively simple life cycle that is easy to control (Shpigel et al. 1999, Neori et al. 2004, Robertson-Andersson et al. 2006, 2008). Seaweeds such as *U. lactuca* that are co-cultured with abalone in an integrated aquaculture system therefore add considerable value to aquaculture because not only do they serve as biological filters that reduce the filtration costs by recycling the nutrients within the system, but they are a potential high-protein feed (Shpigel et al. 1999, Neori et al. 2003, 2004, Troell et al. 2003, 2006, Robertson-Andersson et al. 2006, 2008, Nobre et al. 2010).

In conclusion, this study is the first to document the effect of farm-grown, protein-enriched *U. lactuca* on the growth of the abalone *H. midae*. *Ulva lactuca* grown in aquaculture effluent produced significantly better growth in *H. midae* than their wild

counterparts. While the protein-enriched *U. lactuca* on its own was not a good substitute for kelp in this study, when fed in combination with kelp as a mixed diet, however, it produced the best growth in *H. midae*. The potential for on-farm cultivation of *U. lactuca* may become even more important as many kelp beds have approached limits of sustainable harvesting, especially in kelp concession areas with high numbers of abalone farms (Anderson et al. 2003, 2006, Rothman et al. 2006, Troell et al. 2006). Furthermore, farms that feed only formulated feeds may at some stage have to feed macroalgal diets to reduce their overall FFER¹. In light of these challenges, those farms with reduced access to abundant fresh kelp and those located substantial distances away from any kelp resource, as well as those that rely heavily on formulated feed, may find it necessary to resort to on-farm cultivation of macroalgae such as *U. lactuca*. Co-culturing *U. lactuca* alongside *H. midae* in integrated aquaculture systems clearly hold benefits for the South African abalone farming industry.

Acknowledgements

We thank the Department of Biodiversity and Conservation Biology (BCB) at the University of the Western Cape, and the Jacobsbaai Sea Products commercial abalone farm for providing funding, research facilities and technical support. We are grateful to the South African National Research Foundation (NRF) and the Swedish International Development Agency (SIDA) for providing scholarships as well as running expenses for this research. We are grateful to Peter Britz (Rhodes University) for his continued support and most valuable comments. Special thanks to Vanessa Couldridge (BCB) for assistance with the statistical analyses of the data. Two anonymous reviewers are gratefully thanked for their contributions in strengthening the manuscript.

¹Feed Fish Equivalency Ratio (FFER, also referred to as Forage Fish Efficiency Ratio) is defined by the WWF (<http://www.worldwildlife.org/what/globalmarkets/aquaculture/feedfaq.html#2>) as a means of estimating the amount of wild forage fish used to produce a unit of farmed fish. The calculation takes into account the yield of forage fish live weight to dry fishmeal weight, the efficiency of feed use (the feed conversion ratio or FCR) and the inclusion rates of fishmeal and fish oil in feed. In the context of the Aquaculture Dialogues (“fishmeal” debates), FFER rather than FCR is being considered as a potential indicator for individual farm performance.

References

- Anderson RJ, Bolton JJ, Molloy FJ, Rotmann KWG. 2003. Commercial seaweeds in southern Africa. In: Chapman ARO, Anderson RJ, Vreeland VJ, Davison IR (eds), *Proceedings of the 17th International Seaweed Symposium*. Oxford: Oxford University Press. pp 1-12.
- Anderson RJ, Rothman MD, Share A, Drummond H. 2006. Harvesting of the kelp *Ecklonia maxima* in South Africa affects its three obligate, red algal epiphytes. *Journal of Applied Phycology* 18: 343-349.
- Barkai R, Griffiths CL. 1988. Diet of the South African abalone *Haliotis midae*. *South African Journal of Marine Science* 4: 37-44.
- Bautista-Teruel MN, Millamena OM. 1999. Diet development and evaluation for juvenile abalone, *Haliotis asinina*: protein/energy levels. *Aquaculture* 178: 117-126.
- Bautista-Teruel MN, Fermin AC, Koshio SS. 2002. Diet development and evaluation for juvenile abalone, *Haliotis asinina*: animal and plant protein sources. *Aquaculture* 219: 645-653.
- Bolton JJ. 2006. Do we have the vision to integrate our marine aquaculture? *South African Journal of Science: Commentary* 102: 507-508.
- Bolton JJ, Robertson-Andersson DV, Shuuluka D, Kandjengo L. 2009. Growing *Ulva* (Chlorophyta) in integrated systems as a commercial crop for abalone feed in South Africa: a SWOT analysis. *Journal of Applied Phycology* 21: 575-583.
- Britz PJ. 1995. The nutritional requirements of *Haliotis midae* and development of a practical diet for intensive abalone culture. PhD thesis, Rhodes University, South Africa.
- Britz PJ. 1996a. Effects of dietary protein level on growth performance of South African abalone, *Haliotis midae*, fed on fishmeal based semi-purified diets. *Aquaculture* 140: 55-61.
- Britz PJ. 1996b. The suitability of selected protein sources for inclusion in formulated diets for the South African abalone *Haliotis midae*. *Aquaculture* 140: 63-73.
- Britz PJ, Hecht T. 1997. Effect of dietary protein and energy level on growth and body composition of South African abalone, *Haliotis midae*. *Aquaculture* 156: 195-210.
- Cook PA, Claydon C. 1991. Preliminary assessment of growth rates of farmed perlemoen. In: Heath RGM (ed), *Aquaculture '90. Proceedings of a Symposium of the Aquaculture Association of South Africa*. pp 105-109.
- Day RW, Fleming AE. 1992. The determinants and measurement of abalone growth. Abalone of the World, Biology, Fisheries and Culture. In: Shepherd SA, Tegner M J, Guzmán Del Prío (Eds). *Abalone of the World, Biology, Fisheries and Culture*. Oxford: Blackwell Scientific Publications. pp 169-181.

- Demetropoulos CL, Langdon CJ, 2004a. Pacific dulse (*Palmaria mollis*) as a food and biofilter in recirculated, land-based abalone culture systems. *Aquacultural Engineering* 32: 57-75.
- Demetropoulos CL, Langdon CJ. 2004b. Effects of nutrient enrichment and biochemical composition of diets of *Palmaria mollis* on growth and condition of Japanese abalone, *Haliotis discus hannai* and red abalone, *Haliotis rufescens*. *Journal of Experimental Marine Biology and Ecology* 308: 185-206.
- Dixon MG. 1992. The effect of temperature and photoperiod on the digestive physiology of *Haliotis midae*. MSc. thesis, Rhodes University, South Africa.
- Dlaza TS, Maneveldt GW, Viljoen C. 2008. The growth of post-weaning abalone *Haliotis midae* (Linnaeus) fed commercially available formulated feeds supplemented with fresh wild seaweed. *African Journal of Marine Science* 30: 199-203.
- Erasmus JH, Cook PA, Coyne VE. 1997. The role of bacteria in the digestion of seaweed by the abalone *Haliotis midae*. *Aquaculture* 155: 377-386.
- Fleming AE. 1995a. Growth, intake, feed conversion efficiency and chemosensory preference of the Australian abalone *Haliotis rubra*. *Aquaculture* 132: 297-311.
- Fleming AE. 1995b. Digestive efficiencies of the Australian blacklip abalone *Haliotis rubra* in relation to growth and feed preference. *Aquaculture* 34: 279-293.
- Fleming RJ, van Barneveld PW, Hone PW. 1996. The development of artificial diets for abalone: a review and future directions. *Aquaculture* 140: 5-53.
- Fleurence J, Chenard E, Lucon M. 1999. Determination of the nutritional value of proteins obtained from *Ulva armoricana*. *Journal of Applied Phycology* 11: 231-239.
- Francis TL, Maneveldt GW, Venter J. 2008a. Determining the most appropriate feeding regime for the South African abalone *Haliotis midae* Linnaeus grown on kelp. *Journal of Applied Phycology* 20: 597-602.
- Francis TL, Maneveldt GW, Venter J. 2008b. Growth of market-size abalone (*Haliotis midae*) fed kelp (*Ecklonia maxima*) versus a low-protein commercial feed. *African Journal of Aquatic Science* 33: 279-282.
- Green AJ, Jones CLW, Britz PJ. 2011. The protein and energy requirements of farmed South African abalone *Haliotis midae* cultured at optimal and elevated water temperatures. *Aquaculture Research* 42: no. doi:10.1111/j.1365-2109.2010.02759.x.
- Hahn KO. 1989. Nutrition and growth of abalone. In: Hahn KO (ed), *CRC Handbook of Culture of Abalone and other marine gastropods*. Boca Raton, Florida: CRC Press. pp 135-156.
- Hattingh A. 2006. The effect of three Abfeed diets, fresh kelp and a combination diet on FCR, growth rates and meat yield of *Haliotis midae*. Marifeed (Pty) Ltd

- Research and Development, <http://www.abfeed.com/mari-resdev-completed.htm>. Accessed 01 February 2011.
- Kruatrachue MS, Sawatpeera ES, Chitramyong Y, Sonchaeng P, Upatham ES, Sangpradub S. 2004. Comparative growth performance of early juvenile *Haliotis asinine* fed various artificial diets. *Journal of Shellfish Research* 23: 197-204.
- Lee SM. 2004. Utilization of dietary protein, lipid and carbohydrate by abalone *Haliotis discus hannai*: a review. *Journal of Shellfish Research* 23: 1027-1030.
- Mercer JP, Mai K, Donlon J. 1993. Comparative studies on the nutrition of two species of abalone, *Haliotis tuberculata* L. and *Haliotis discus hannai* Ino.: I. Effects of algal diets on growth and biochemical composition. *Invertebrate Reproductive Development* 23: 75-88.
- Naidoo K, Maneveldt G, Ruck K, Bolton JJ. 2006. A comparison of various seaweed-based diets and formulated feed on growth rate of abalone in a land-based aquaculture system. *Journal of Applied Phycology* 18: 437-443.
- Neori A, Shpigel M. 1999. Using algae to treat effluents and feed invertebrates in sustainable integrated mariculture. *Journal of the World Aquaculture Society* 30: 46-49.
- Neori A, Krom MD, Ellner SP, Boyd CE, Popper D, Rabinovitch R, Davison PJ, Dvir O, Zuber D, Ucko M, Angel D, Gordin H. 1996. Seaweed biofilters as regulators of water quality in integrated fish-seaweed culture units. *Aquaculture* 141: 183-199.
- Neori A, Ragg NLC, Shpigel M. 1998. The integrated culture of seaweed, abalone, fish and clams in modular intensive land-based systems: II. Performance and nitrogen partitioning within an abalone (*Haliotis tuberculata*) and macroalgae culture system. *Aquacultural Engineering* 17: 215-239.
- Neori A, Msuya FE, Shauli L, Schuenhoff A, Kopel F, Shpigel M. 2003. A novel three stage seaweed (*Ulva lactuca*) biofilter design for integrated mariculture. *Journal of Applied Phycology* 15: 543-553.
- Neori A, Chopin T, Troell M, Buschmann AH, Kraemer GP, Halling C, Shpigel M, Yarish C. 2004. Integrated aquaculture: rationale, evolution and state of the art emphasizing seaweed biofiltration in modern mariculture. *Aquaculture* 231: 361-391.
- Nobre A.M, Neori A, Robertson-Andersson D, Sankar K. 2010. Ecological-economic assessment of aquaculture options: Comparison between abalone monoculture and integrated multi-trophic aquaculture of abalone and seaweeds. *Aquaculture* 306: 116-126.
- Owen B, Disaivo LH, Ebert EE, Fonck E. 1984. Culture of the California red abalone *Haliotis rufescens* Swanson in Chile. *Veliger* 27: 101-105.
- Robertson-Andersson DV, Leitao D, Bolton JJ, Anderson RJ, Njobeni A, Ruck K. 2006. Can kelp extract (KELPAK™) be useful in seaweed mariculture? *Journal of Applied Phycology* 18: 315-321.

- Robertson-Andersson DV, Potgieter M, Hansen J, Bolton JJ, Troell M, Anderson RJ, Halling C, Probyn T. 2008. Integrated seaweed cultivation on an abalone farm in South Africa. *Journal of Applied Phycology* 20: 579-595.
- Rosen G, Langdon CJ, Evans F. 2000. The nutritional value of *Palmaria mollis* cultured under different light intensities and water exchange rates for juvenile red abalone *Haliotis rufescens*. *Aquaculture* 185: 121-136.
- Rothman MD, Anderson RJ, Smit AJ. 2006. The effects of harvesting of the South African kelp (*Ecklonia maxima*) on kelp population structure growth rate and recruitment. *Journal of Applied Phycology* 18: 335-341.
- Sales J, Truter PJ, Britz PJ. 2003a. Optimum dietary crude protein level for growth in South African abalone (*Haliotis midae* L.). *Aquaculture Nutrition* 9: 85-89.
- Sales J, Britz PJ, Viljoen J. 2003b. Dietary phosphorus leaching and apparent phosphorus digestibility from different inorganic phosphorus sources for South African abalone (*Haliotis midae* L.). *Aquaculture Nutrition* 9: 169-174.
- Shpigel M, Neori A. 1996. Abalone and seaweeds intensive cultivation in integrated land-based mariculture system. I. Proposed design and cost analyses. *Aquacultural Engineering* 15: 313-26.
- Shpigel M, Marshall A, Lupatsch I, Mercer JP, Neori A. 1996. Acclimation and Propagation of the Abalone *Haliotis tuberculata* in a Land-Based Culture System in Israel. *Journal of the World Aquaculture Society* 27: 435-442.
- Shpigel M, Ragg NL, Lupatsch I, Neori A. 1999. Protein Content Determines the Nutritional Value of the Seaweed *Ulva lactuca* L. for the Abalone *Haliotis tuberculata* L. and *H. discus hannai* Ino. *Journal of Shellfish Research* 18: 227-33.
- Simpson JA, Cook PA. 1998. Rotation Diets: A method of improving growth of cultured abalone using natural algal diets. *Journal of Shellfish Research* 17: 635-640.
- Stepito NK, Cook PA. 1993. Feeding preferences of the juvenile South African abalone *Haliotis midae* (Linnaeus, 1758). *Journal of Shellfish Research* 15: 653-657.
- Tahil AS, Juinio-Menez MA. 1999. Natural diet, feeding periodicity and functional response to food density of the abalone, *Haliotis asinina* L. (Gastropoda). *Aquaculture Research* 30: 95-107.
- Troell M, Halling C, Neori A, Chopin T, Buschmann AH, Kautsky N, Yarish C. 2003. Integrated mariculture: asking the right questions. *Aquaculture* 226: 69-90.
- Troell M, Robertson-Andersson DV, Anderson RJ, Bolton JJ, Maneveldt G, Halling C, Probyn T. 2006. Abalone farming in South Africa: An overview with perspectives on kelp resources, abalone feed, potential for on-farm seaweed production and socio-economic importance. *Aquaculture* 257: 266-281.

- Viana MT, López LM, Salas A. 1993. Diet development for juvenile abalone *Haliotis fulgens*: Evaluation of two artificial diets and macroalgae. *Aquaculture* 117: 49-156.
- Zar JH. 1984. *Biostatistical Analysis*. Prentice-Hall, Sydney, 718 pp.
- Zhu W, Mai K, Wu G. 2002. Thiamine requirement of juvenile abalone, *Haliotis discus hannai* Ino. *Aquaculture* 207: 331-343.

Table 1. Water quality variables of the experimental system under investigation.

Variable	Mean \pm standard deviation	Range
Salinity (‰)	34 ± 0.91	29 – 40
Temperature (°C)	15.5 ± 2.50	9.8 – 19.2
pH	8.24 ± 0.22	7.1 – 9.1
Dissolved O ₂ (mg L ⁻¹)	8.51 ± 0.67	6.7 – 10.3
Flow rate (L h ⁻¹)	700 ± 100	850 – 1300

Table 2: Proximal analysis (based on dry weights) of the feeds tested in this study. Feed samples were sent to the Animal Production Laboratory, Institute for Animal Production, Department of Agriculture: Western Cape, Elsenberg for nutritional analyses. Comparative values with the same superscript within a column are not statistically different (ANOVA; $p \geq 0.05$).

Feed	Dry Matter (%)	Ash (%)	Protein (%)	Fibre (%)	Fat (%)	Carbohydrate (%)	Gross Energy (MJ kg ⁻¹)
Kelp	88.43 ± 0.45 ^a	26.71 ± 1.96 ^a	9.73 ± 0.68 ^a	8.35 ± 0.14 ^b	0.66 ± 0.13 ^b	62.90 ± 1.15 ^b	10.94 ± 0.20 ^b
<i>U. lactuca</i> (farmed)	88.36 ± 0.98 ^{ab}	23.11 ± 0.71 ^a	25.57 ± 1.45 ^c	6.31 ± 0.44 ^a	0.29 ± 0.04 ^a	51.03 ± 1.68 ^a	13.12 ± 0.27 ^c
<i>U. lactuca</i> (wild)	86.17 ± 0.47 ^b	31.20 ± 0.35 ^b	20.22 ± 0.56 ^b	5.74 ± 0.09 ^a	0.19 ± 0.01 ^a	48.40 ± 0.39 ^a	10.55 ± 0.01 ^a

Table 3: Growth parameters of abalone fed the various diets. Specific growth rate (SGR, % body weight day⁻¹), daily increment in shell length (DISL, $\mu\text{m day}^{-1}$), feed conversion ratio (FCR) and condition factor (CF) are provided for all feeds. Values with the same superscript within a column are not statistically different from each other (ANOVA; $p \geq 0.05$).

Diet treatment	Mean Final Weight (g)	Mean Final Length (mm)	SGR	DISL	FCR	Initial CF	Final CF	SGR	DISL	CF
----------------	-----------------------	------------------------	-----	------	-----	------------	----------	-----	------	----

Kelp + farmed <i>U. lactuca</i>	24.308 ± 0.28 ^a	50.521 ± 0.21 ^a	0.423 ± 0.02 ^a	59.593 ± 0.02 ^a	N/A	1.073 ^a	1.093 ^a	1	1	1
Kelp	21.194 ± 0.28 ^b	48.955 ± 0.23 ^b	0.367 ± 0.02 ^b	53.148 ± 0.02 ^b	47 ^a	1.072 ^a	1.047 ^b	2	2	2
<i>U. lactuca</i> (farmed)	17.568 ± 0.26 ^c	46.486 ± 0.22 ^c	0.290 ± 0.02 ^c	42.988 ± 0.03 ^c	91 ^b	1.072 ^a	1.013 ^c	3	3	3
<i>U. lactuca</i> (wild)	7.164 ± 0.09 ^d	36.950 ± 0.15 ^d	-0.079 ± 0.01 ^d	3.745 ± 0.02 ^d	-71 ^c	1.073 ^a	0.821 ^d	4	4	4

Figure legends

Figure 1: Mean increase in shell length (\pm standard error) of abalone fed kelp, *U. lactuca*, and a combination of the two macroalgae (ANOVA; $p < 0.001$); “F” refers to farm-grown, protein-enriched *U. lactuca*; “W” refers to wild *U. lactuca*.

Figure 2: Mean increase in body weight (\pm standard error) of abalone fed kelp, *U. lactuca*, and a combination of the two macroalgae (ANOVA; $p < 0.001$); “F” refers to farm-grown, protein-enriched *U. lactuca*; “W” refers to wild *U. lactuca*.

Figure 1.

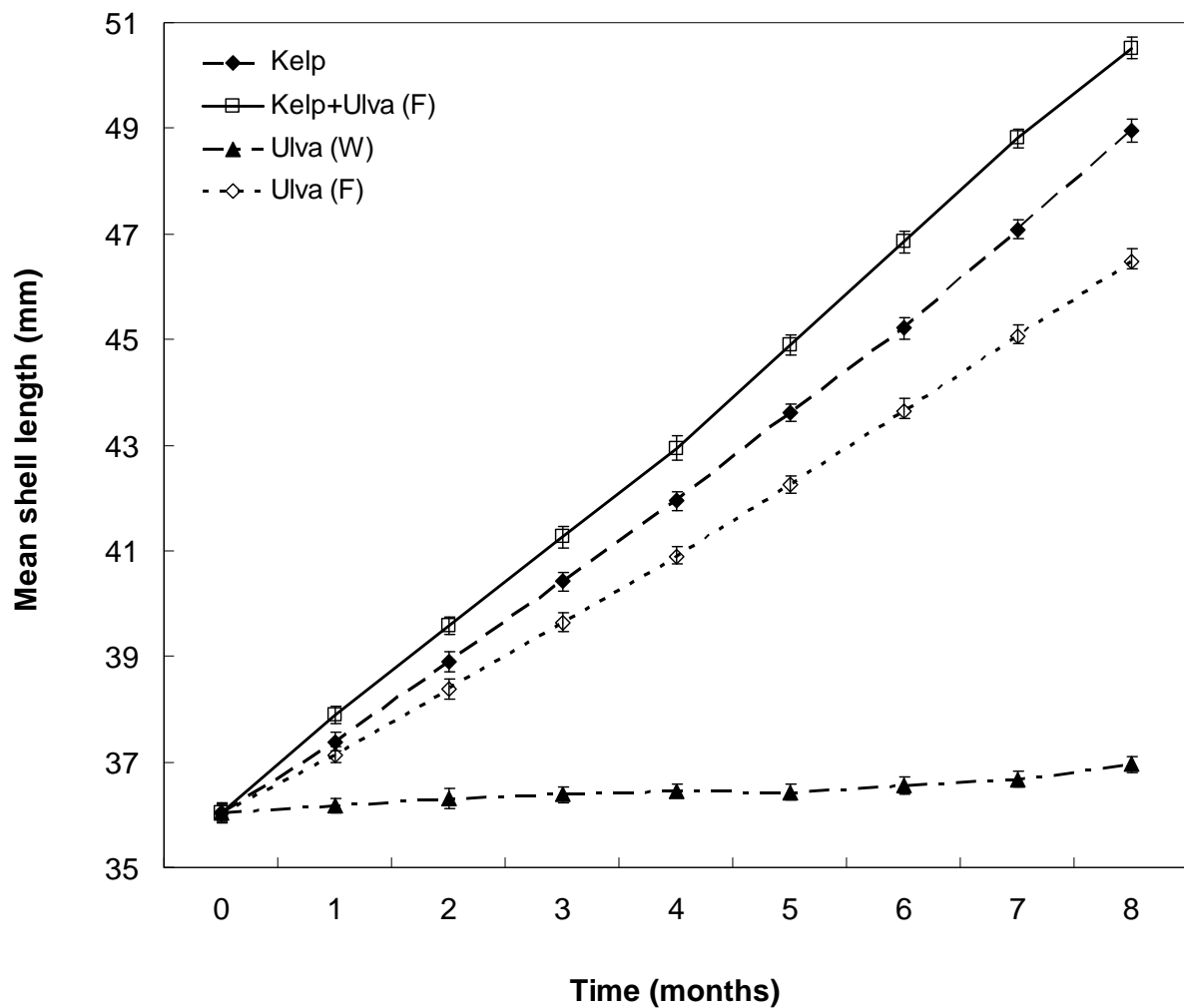


Figure 2.

